Safety Testing of Drug Metabolites

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Safety Testing of Metabolites Guidances Chronology

- Metabolites in Safety Testing (MIST)

  PhRMA Position Paper, Baille et al

  Toxicology and Applied Pharmacology, April 2002

- Safety Testing of Drug Metabolites

  CDER Draft Guidance

  Published in Federal Register, June 2005.

Safety Testing of Metabolites Guidance Rationale

- Independent safety studies are recommended for major human metabolites that are unique (or disproportionately produced) when safety is not adequately assessed in the standard toxicology studies.
- Recommendation applies to " major" metabolites in human plasma that account for > 10% of total systemic exposure.
- Intended for small molecule therapeutics (non-biologics)
- Safety testing generally not required for Phase II metabolites.

Points to Consider

- In vitro and in vivo drug metabolism studies are useful for early decisions regarding the adequacy of the preclinical test species to evaluate the safety of drug metabolites.
- Quantitative and qualitative differences in metabolite profiles are important when comparing exposures in the nonclinical species relative to humans.
- In vivo comparative pharmacokinetics data for the metabolite are considered the most relevant and should be provided as early as possible.
- Qualitative differences in metabolism are extremely rare, i.e., identification of totally unique human metabolites.
- Quantitative differences where a metabolite is produced to a significantly greater extent in humans than in animals is more common. However, the need for independent toxicity testing of major human metabolites is still infrequent.

General Recommendations for Metabolite Safety Testing as per MIST and CDER Guidances

- Genotoxicity – in vitro tests for point mutations and chromosome aberrations

- General Toxicity –

  Testing in a single species (rat) is generally considered adequate

  Duration dependent upon the duration of clinical use – e.g., 2 weeks – 13 weeks

- Embryofetal development - testing with the metabolite in a single species when the metabolite is not produced sufficiently in rats or rabbits dosed with the parent drug

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Genotoxicity Testing

- If the major human metabolite(s) of concern are not produced to any significant degree in the rat, genotoxicity testing employing a rat liver S9 metabolizing system may not adequately test the genotoxic potential of the human metabolites.
- In vitro assays with the metabolite to detect point mutation and chromosomal aberrations are recommended.
- If major metabolites test positive for genotoxicity , additional testing may be necessary. (e.g. in vivo genotoxicity and/or carcinogenicity testing).

Case Example # 1 - Common

- Quantitative differences in metabolism
- Metabolite represents 1-2% of total activity in plasma of rats, 5% in dogs, 20% in humans . Therefore , a major metabolite in human.
- Due to testing of much higher doses in the rat and dog toxicology studies than in human clinical trials, steady state AUC of metabolite in rat and dog >> steady state AUC for metabolite in humans with the maximum recommended human dose (MRHD).
- Therefore, general toxicity studies adequately characterize the toxicity of metabolite in rats and dogs.
- Despite differential metabolism in rats, the concentrations of the major human metabolite in the genotoxicity, rat embryofetal toxicity, and carcinogenicity studies provide adequate exposures and characterization of the major human metabolite.
- No additional testing required.

Case Study # 2 – Rare

- Disproportionate human metabolite requiring extensive safety testing
- Two primary hydroxylated metabolites (M1 and M2) and two secondary oxidative metabolites (M3 and M4) observed in vitro using microsomes and hepatocytes from human, monkey, rat, dog, and mouse liver.
- Human, monkey and dog microsomes metabolized predominantly to M1 and M4, while rat and mouse preferentially formed M2 and M3.

In vivo Metabolism

- Humans produce a secondary oxidative metabolite , M4 at concentrations up to 4-fold higher than parent drug.
- M4 not produced to any significant degree in rats or mice.
- M 4 produced in monkeys but to a lesser degree than in humans.

Steady state AUC_{0-24} of Parent Drug and M4 Metabolite

	Human (MRHD)	Monkey	Rat
Parent	1,800	15,000	12,500
M4	7,700 (80%)	5,000 (25%)	135 (1%)

M 4 Characterization

- Severe class-related toxicity and novel toxicities/target organs not typically seen with the drug class were observed in monkeys.
- M4 not pharmacologically active at the target receptor.
- At highest doses evaluated in monkeys, M4 concentrations were ~ 60% of human exposures with MRHD.
- No additional toxicological characterization of M4 proposed by the sponsor.

CDER Recommendations for Toxicological Characterization of M4

As per the MIST White Paper and CDER Draft Guidance the following studies with M4 metabolite were recommended :

In vitro genotoxicity testing

3-month toxicity study in rats

A rat embryofetal development study
(unless rabbits produce adequate M4)

Completion of these studies prior to Phase 3 trials was recommended

Genetic Toxicology Results

- Parent drug tested negative for genotoxic potential in the standard ICH test battery.
- Metabolite M4 tested positive in the Ames and in vitro chromosomal aberrations assays.
- Division and Executive Carcinogenicity Assessment Committee (ECAC) recommended that an evaluation of the carcinogenic potential of M4 be conducted since M4 not produced to any significant extent in mice or rats.

Carcinogenicity Testing

- Division recommended addition of M4 dose groups or spiking of parent drug groups with M4 in the rat carcinogenicity study.
- Sponsor proposed use of doses 1 and 25X AUC of M4 with the maximum recommended human dose.
- M4 genotoxic so can’t use 25X human AUC ratio for dose selection.
- Three month rat general toxicity study with M4 delayed, so MTD for M4 metabolite was unknown.

Carcinogenicity Testing (cont.)

- Sponsor proposed a P53 transgenic mouse assay (to be submitted prior to Phase 3) and an independent rat carcinogenicity study with M4.
- Rat carcinogenicity study with M4 metabolite delayed until 13-week toxicity data available for dose selection based on MTD.
- Division agreed to accept an NDA submission which included the results of rat and mouse carcinogenicity studies for the parent drug and a P53 transgenic assay with M4.
- The 2-year rat carcinogenicity study with M4 to be provided as soon as available. (during NDA review cycle).

Conclusions

- Absence of pharmacological activity at the target receptor does not eliminate the need for toxicological qualification of a unique or disproportionate major human metabolite.
- When the unique/disproportionate human metabolite is produced at clinical exposures only in the non-rodent species, general toxicity is adequately characterized. However, genotoxicity, reproductive toxicity, and carcinogenicity may not be since those studies are conducted in rodents.
- In some cases, more extensive toxicological characterization of a “ unique” major human metabolite than is recommended by the MIST or CDER guidances may be necessary.

Conclusions

- Under some circumstances safety testing of metabolites may not be necessary (e.g., metabolite of a cytotoxic drug for an oncology indication).

  Sponsors can submit a scientific justification to support a waiver of metabolite safety testing when the weight of evidence suggests a minimal safety concern (negative SAR, similarity to parent drug, very low exposures, etc).